

WHAT IS CLAIMED IS:

1. A method of inhibiting HCV replication in an HCV infected cell comprising the step of providing to said cell an effective amount of a compound
 5 that inhibits NS2/3 autocleavage.

2. The method of claim 1, wherein said compound is selected from the group consisting of:
 an HCV inhibitor polypeptide comprising an NS4A fragment at
 10 least about 11 amino acids in length, wherein said fragment can inhibit autocleavage of NS2/3;
 a pharmaceutically acceptable salt of said HCV inhibitor polypeptide; and
 a prodrug thereof.

3. The method of claim 1, wherein compound is selected from the group consisting of:
 a polypeptide having the structure:

20 $Z^1-Y^1_m-X^1X^2X^3X^4X^5GX^6X^7X^8X^9X^{10}-Y^2_n-Z^2$

wherein X^1 is either serine, cysteine, or threonine;
 X^2 is either valine, leucine, or isoleucine;
 X^3 is either valine, leucine, isoleucine, serine, cysteine or threonine;
 25 X^4 is either valine, leucine, or isoleucine;
 X^5 is either valine, leucine, or isoleucine;
 X^6 is either lysine, arginine, or histidine;
 X^7 is either valine, leucine, or isoleucine;
 X^8 is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine,
 30 or histidine;
 X^9 is either valine, leucine, or isoleucine;
 X^{10} is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;
 each Y^1 is an independently selected amino acid,
 35 each Y^2 is an independently selected amino acid,

Z¹ is an optionally present protecting group covalently joined to Y¹,
 Z² is an optionally present protecting group covalently joined to Y²,
 m is from 0 to 300, and
 n is from 0 to 300,

5 a pharmaceutically acceptable salt of said polypeptide; and
 a prodrug thereof.

4. The method of claim 3, wherein m is from 0 to 25 and n is from
 0 to 25.

10 5. The method of claim 4, wherein said compound is said
 polypeptide or a pharmaceutically acceptable salt thereof.

6. The method of claim 1, wherein said compound is selected
 15 from the group consisting of:
 KGSVVIVGRILSGRK (SEQ. ID. NO. 16),
 Ac-GGSVVIVGRILSGRK (SEQ. ID. NO. 18),
 GGSVVIVGRILSGRG (SEQ. ID. NO. 19),
 KKGSVVIVGRILSGRPAIVPRR-NH₂ (SEQ. ID. NO. 20), and
 20 KKGSVVIVGRILSGRPAIVPDRELLYQEFDE (SEQ. ID. NO. 21),
 or a pharmaceutically acceptable salt thereof.

7. A method of inhibiting HCV replication in an HCV infected
 cell comprising the step of introducing into said cell an effective amount of a nucleic
 25 acid comprising a nucleotide sequence encoding for a polypeptide comprising an
 NS4A fragment at least about 11 amino acids in length, wherein said fragment
 inhibits autocleavage of NS2/3.

8. The method of claim 7, wherein said nucleic acid is an
 30 expression vector.

9. A method of inhibiting HCV replication in an HCV infected
 cell comprising the step of introducing into said cell an effective amount of a nucleic

acid comprising a nucleotide sequence encoding for a polypeptide having the structure:



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wherein X^1 is either serine, cysteine, or threonine;

X^2 is either valine, leucine, or isoleucine;

X^3 is either valine, leucine, isoleucine, serine, cysteine or threonine;

X^4 is either valine, leucine, or isoleucine;

10 X^5 is either valine, leucine, or isoleucine;

X^6 is either lysine, arginine, or histidine;

X^7 is either valine, leucine, or isoleucine;

X^8 is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine, or histidine;

15 X^9 is either valine, leucine, or isoleucine;

X^{10} is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;

each Y^1 is an independently selected amino acid;

each Y^2 is an independently selected amino acid;

20 m is from 0 to 300; and

n is from 0 to 300.

10. The method of claim 9, wherein said nucleic acid is an expression vector.

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11. The method of claim 10, wherein m is from 0 to 25, and n is from 0 to 25.

12. A method of treating a patient for HCV comprising the step of inhibiting NS2/3 autocleavage.

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13. The method of claim 12, wherein said patient is a human patient and said method further comprises the step of identifying said patient as infected with HCV prior to said inhibiting.

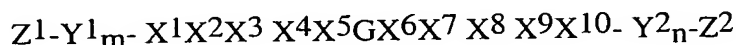
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14. The method of claim 12, wherein said step of inhibiting NS2/3 autocleavage is achieved using an effective amount of a compound selected from the group consisting of:

- a polypeptide comprising an NS4A fragment at least about 11 amino acids in length;
- a pharmaceutically acceptable salt of said polypeptide; and
- a prodrug thereof.

15. The method of claim 12, wherein said step of inhibiting NS2/3 autocleavage is achieved using an effective amount of a compound selected from the group consisting of:

a polypeptide having the structure:



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wherein X^1 is either serine, cysteine, or threonine;

X^2 is either valine, leucine, or isoleucine;

X^3 is either valine, leucine, isoleucine, serine, cysteine or threonine;

X^4 is either valine, leucine, or isoleucine;

20 X^5 is either valine, leucine, or isoleucine;

X^6 is either lysine, arginine, or histidine;

X^7 is either valine, leucine, or isoleucine;

X^8 is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine, or histidine;

25 X^9 is either valine, leucine, or isoleucine;

X^{10} is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;

each Y^1 is an independently selected amino acid,

each Y^2 is an independently selected amino acid,

30 Z^1 is an optionally present protecting group covalently joined to Y^1 ,

Z^2 is an optionally present protecting group covalently joined to Y^2 ,

m is from 0 to 300, and

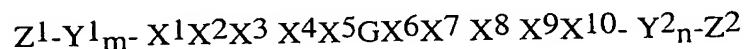
n is from 0 to 300,

- a pharmaceutically acceptable salt of said polypeptide; and
- a prodrug thereof.

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16. A method of inhibiting or preventing HCV replication in a patient comprising the step of treating said patient with an effective amount of a compound selected from the group consisting of:

- 5 a polypeptide that either comprises an NS4A fragment at least about 11 amino acids in length able to inhibit NS2/3 autocleavage or has the structure:



- 10 wherein X^1 is either serine, cysteine, or threonine;
 X^2 is either valine, leucine, or isoleucine;
 X^3 is either valine, leucine, isoleucine, serine, cysteine or threonine;
 X^4 is either valine, leucine, or isoleucine;
 X^5 is either valine, leucine, or isoleucine;
15 X^6 is either lysine, arginine, or histidine;
 X^7 is either valine, leucine, or isoleucine;
 X^8 is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine, or histidine;
 X^9 is either valine, leucine, or isoleucine;
20 X^{10} is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;
each Y^1 is an independently selected amino acid,
each Y^2 is an independently selected amino acid,
 Z^1 is an optionally present protecting group covalently joined to Y^1 ,
25 Z^2 is an optionally present protecting group covalently joined to Y^2 ,
m is from 0 to 300, and
n is from 0 to 300,

a pharmaceutically acceptable salt of said polypeptide; and
a prodrug thereof.

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17. The method of claim 16, wherein said patient is a human infected with HCV.

18. The method of claim 17, wherein said compound is said
35 polypeptide or a pharmaceutically acceptable salt thereof.

19. The method of claim 17, wherein said compound is selected from the group consisting of:

KGSVVIVGRILSGRK (SEQ. ID. NO. 16),

5 Ac-GGSVVIVGRILSGRK (SEQ. ID. NO. 18),

GGSVVIVGRILSGRG (SEQ. ID. NO. 19),

KKGSVVIVGRILSGRPAIVPRR-NH₂ (SEQ. ID. NO. 20), and

KKGSVVIVGRILSGRPAIVPDRELLYQEFDE (SEQ. ID. NO. 21),

or a pharmaceutically acceptable salt thereof.

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20. A method of inhibiting or preventing HCV replication in a patient comprising the step of administering to said patient an effective amount of a nucleic acid comprising a nucleotide sequence encoding for a polypeptide comprising an NS4A fragment at least about 11 amino acids in length, wherein said fragment

15 inhibits autocleavage of NS2/3.

21. The method of claim 20, wherein said nucleic acid is an expression vector.

20 22. A method of inhibiting or preventing HCV replication in a patient comprising the step of administering to said patient an effective amount of a nucleic acid comprising a nucleotide sequence encoding for a polypeptide having the structure:

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$$Y^1_m - X^1 X^2 X^3 X^4 X^5 G X^6 X^7 X^8 X^9 X^{10} - Y^2_n$$

wherein X¹ is either serine, cysteine, or threonine;

X² is either valine, leucine, or isoleucine;

X³ is either valine, leucine, isoleucine, serine, cysteine or threonine;

30 X⁴ is either valine, leucine, or isoleucine;

X⁵ is either valine, leucine, or isoleucine;

X⁶ is either lysine, arginine, or histidine;

X⁷ is either valine, leucine, or isoleucine;

X⁸ is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine,

35 or histidine;

X⁹ is either valine, leucine, or isoleucine;

X¹⁰ is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;

each Y¹ is an independently selected amino acid;

5 each Y² is an independently selected amino acid;

m is from 0 to 300; and

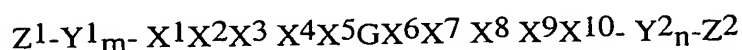
n is from 0 to 300.

10 23. The method of claim 22, wherein said nucleic acid is an expression vector.

24. The method of claim 22, wherein m is from 0 to 25 and n is from 0 to 25.

15 25. A compound selected from the group consisting of:
a pharmaceutically acceptable salt of a HCV inhibitor polypeptide, wherein said HCV inhibitor polypeptide comprises an NS4A fragment at least about 11 amino acids in length and can inhibit autocleavage of NS2/3; and
a prodrug thereof.

20 26. A compound selected from the group consisting of:
a polypeptide having the structure:



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wherein X¹ is either serine, cysteine, or threonine;

X² is either valine, leucine, or isoleucine;

X³ is either valine, leucine, isoleucine, serine, cysteine or threonine;

X⁴ is either valine, leucine, or isoleucine;

30 X⁵ is either valine, leucine, or isoleucine;

X⁶ is either lysine, arginine, or histidine;

X⁷ is either valine, leucine, or isoleucine;

X⁸ is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine, or histidine;

X⁹ is either valine, leucine, or isoleucine;

X¹⁰ is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;

each Y¹ is an independently selected amino acid,

5 each Y² is an independently selected amino acid,

Z¹ is an optionally present protecting group covalently joined to Y¹,

Z² is an optionally present protecting group covalently joined to Y²,

m is from 0 to 300, and

n is from 0 to 300;

10 a pharmaceutically acceptable salt of said polypeptide; and
a prodrug thereof;

provided that if said compound is said polypeptide then at least one of Z¹ or Z² is present.

15 27. The compound of claim 26, wherein m is from 0 to 25, and n is from 0 to 25.

28. The compound of claim 27, wherein said compound is said pharmaceutically acceptable salt.

20 29. A compound selected from the group consisting of:

KGSVVIVGRILSGRK (SEQ. ID. NO. 16),

Ac-GGSVVIVGRILSGRK (SEQ. ID. NO. 18),

GGSVVIVGRILSGRG (SEQ. ID. NO. 19),

25 KKGSVVIVGRILSGRPAIVPRR-NH₂ (SEQ. ID. NO. 20), and

KKGSVVIVGRILSGRPAIVPDRELLYQEFDE (SEQ. ID. NO. 21),

or a pharmaceutically acceptable salt thereof.

30. A nucleic acid comprising a nucleotide sequence encoding for
30 the HCV inhibitor polypeptide of claim 25.

31. The nucleic acid of claim 30, wherein said nucleic acid is an expression vector.

32. A nucleic acid comprising a nucleotide sequence encoding for the polypeptide of claim 26.

33. The nucleic acid of claim 32, wherein said nucleic acid is an expression vector.

34. A pharmaceutical composition for inhibiting HCV replication comprising;
a pharmaceutically acceptable carrier; and
an effective amount of a compound selected from the group consisting of:
an HCV inhibitor polypeptide comprising an NS4A fragment at least about 11 amino acids in length, wherein said fragment can inhibit autocleavage of NS2/3;
a pharmaceutically acceptable salt of said HCV inhibitor polypeptide; and
a prodrug thereof.

35. A pharmaceutical composition for inhibiting HCV replication comprising:
a pharmaceutically acceptable carrier; and
an effective amount of a polypeptide having the structure:



wherein X¹ is either serine, cysteine, or threonine;
X² is either valine, leucine, or isoleucine;
X³ is either valine, leucine, isoleucine, serine, cysteine or threonine;
X⁴ is either valine, leucine, or isoleucine;
X⁵ is either valine, leucine, or isoleucine;
X⁶ is either lysine, arginine, or histidine;
X⁷ is either valine, leucine, or isoleucine;
X⁸ is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine, or histidine;
X⁹ is either valine, leucine, or isoleucine;

X¹⁰ is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;

each Y¹ is an independently selected amino acid,

each Y² is an independently selected amino acid,

5 Z¹ is an optionally present protecting group covalently joined to Y¹,

Z² is an optionally present protecting group covalently joined to Y²,

m is from 0 to 300, and

n is from 0 to 300;

10 a pharmaceutically acceptable salt of said polypeptide; and
a prodrug thereof.

36. A pharmaceutical composition for inhibiting HCV replication comprising: a pharmaceutically acceptable carrier; and an effective amount of a nucleic acid encoding for a polypeptide comprising a fragment of NS4A at least about
15 11 amino acids in length, wherein said fragment can inhibit autocleavage of NS2/3.

37. The composition of claim 36, wherein said nucleic acid is present in an expression vector providing for expression in a human.

20 38. A pharmaceutical composition for inhibiting HCV replication comprising a pharmaceutically acceptable carrier and an effective amount of a nucleic acid encoding for a polypeptide having the structure:



25 wherein X¹ is either serine, cysteine, or threonine;

X² is either valine, leucine, or isoleucine;

X³ is either valine, leucine, isoleucine, serine, cysteine or threonine;

X⁴ is either valine, leucine, or isoleucine;

30 X⁵ is either valine, leucine, or isoleucine;

X⁶ is either lysine, arginine, or histidine;

X⁷ is either valine, leucine, or isoleucine;

X⁸ is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine, or histidine;

X⁹ is either valine, leucine, or isoleucine;

X¹⁰ is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or glutamic acid;

each Y¹ is an independently selected amino acid;

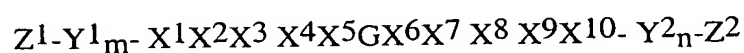
5 each Y² is an independently selected amino acid;

m is from 0 to 300; and

n is from 0 to 300.

39. The composition of claim 38, wherein said nucleic acid is
10 present in an expression vector providing for expression in a human.

40. A method for inhibiting HCV polyprotein processing
comprising the step of contacting a cell expressing an HCV polypeptide that contains
at least NS2/3 with an inhibitory polypeptide that either comprises an NS4A fragment
15 at least about 11 amino acids in length able to inhibit NS2/3 autocleavage or has the
structure:



20 wherein X¹ is either serine, cysteine, or threonine;

X² is either valine, leucine, or isoleucine;

X³ is either valine, leucine, isoleucine, serine, cysteine or threonine;

X⁴ is either valine, leucine, or isoleucine;

X⁵ is either valine, leucine, or isoleucine;

25 X⁶ is either lysine, arginine, or histidine;

X⁷ is either valine, leucine, or isoleucine;

X⁸ is either aspartic acid, glutamic acid, valine, leucine, isoleucine, lysine, arginine,
or histidine;

X⁹ is either valine, leucine, or isoleucine;

30 X¹⁰ is either serine, cysteine, threonine, asparagine, glutamine, aspartic acid, or
glutamic acid;

each Y¹ is an independently selected amino acid,

each Y² is an independently selected amino acid,

Z¹ is an optionally present protecting group covalently joined to Y¹,

Z² is an optionally present protecting group covalently joined to Y²,
 m is from 0 to 300, and
 n is from 0 to 300,

- 5 a pharmaceutically acceptable salt of said inhibitory
 polypeptide; and
 a prodrug thereof.

41. The method of claim 40, wherein said polypeptide is selected
 from the group consisting of:
 10 KGSVVIVGRILSGRK (SEQ. ID. NO. 16),
 Ac-GGSVVIVGRILSGRK (SEQ. ID. NO. 18),
 GGSVVIVGRILSGRG (SEQ. ID. NO. 19),
 KKGSVVIVGRILSGRPAIVPRR-NH₂ (SEQ. ID. NO. 20), and
 KKGSVVIVGRILSGRPAIVPDRELLYQEFDE (SEQ. ID. NO. 21),
 15 or a pharmaceutically acceptable salt thereof.

42. A method of screening for a compound that inhibits HCV
 replication or HCV polyprotein processing comprising the steps of:
 a) selecting for a compound that binds to the NS4A target
 20 site using a polypeptide comprising NS2/3 or a binding portion thereof, and
 b) measuring the ability of said compound to inhibit HCV
 replication or HCV polyprotein processing.

43. The method of claim 42, wherein said method measures the
 25 ability of said compound to inhibit HCV polyprotein processing.

44. The method of claim 42, wherein said step (b) is performed in
 the presence of a non-saturating amount of a NS4A agonist.

- 30 45. A method of screening for a compound that inhibits HCV
 replication or HCV polyprotein processing comprising the step of measuring the
 ability of said compound to inhibit HCV replication or HCV polyprotein processing in
 the presence of a non-saturating amount of a NS4A agonist.